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Biochemical Performance of *Cirrhinus cirrhosus* Fingerlings after Soya Chunk and Almond Supplementation

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Abstract: The purpose of the study was to investigate how *Cirrhinus cirrhosus* fingerlings responded biochemically to the addition of soy chunks and almonds. Fish were divided into four groups-- Group I served as the control group, Group II was given the 1% soya chunk diet I, Group III was given the 1% almond diet II, and Group IV was given the 1% combined diet (1: 1 ratio of Diet I and Diet II). Overall, it can be said that providing *Cirrhinus cirrhosus* with Soya Chunk, Almond, or Soya Chunk + Almond supplements has a considerable impact on the species' biochemical makeup and growth performance. Dietary benefits are directly correlated with the length of supplementation. In comparison to control, other diets, and 15 days of diet supplementation, the combined diet (Soya Chunk + Almond) had a significant impact on growth performance and biochemical composition at 30 days of supplementation. The findings of this study indicated that the optimal nutritional supplements for fingerling *Cirrhinus cirrhosus* is a mixed diet.

Keywords: *Cirrhinus cirrhosus* fingerlings, Soya Chunk, Almond Diet, Combined Diet, Growth performance, Biochemical composition

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Introduction

India is second in aquaculture production. China's one-fifth population harvests one-third of the world's fish and cultivates two-thirds (FAO, 2016). While India uses 3-6 species in a single pond, the Chinese use 10 or more to maximize yield. Freshwater aquaculture has contributed over 95% of India's aquaculture production in the last two decades. India has 3.15 million hectares of

reservoirs, 2.36 million ha of ponds/tanks, and 0.19 million ha of rivers/canals. Freshwater aquaculture's share of inland fisheries increased from 34% in the 1980s to 80% during 2016-17 (DADF, 2017).

Catla (*Catla catla*), rohu (*Labeo rohita*), and mrigal (*Cirrhinus cirrhosus*) are three significant carps with great market value and demand in

India. Due to their taste and flesh, they are the most significant commercial fish in India. They provide fats and proteins (Swapna *et al.*, 2010). They produce 67% of freshwater fish (ICLARM, 2001).

Cirrhinus mrigala contributes significantly to south India's freshwater fish output. Fish farming depends on proper feeding management. The trend in fish culture is towards higher intensification, which requires feeds. Growth depends on well-balanced, nutritionally full, and cost-effective compounded feeds. Fish need food to grow and live. Fish devour plants and animals in nature. In semi-intensive fish culture, artificial feed is used to sustain a higher fish density than the water's natural fertility can support (Jhingran, 1991). This study examined the biochemical performance of *Cirrhinus cirrhosis* fingerlings supplemented with Soya Chunks and Almonds.

Materials and Methods

Fish collection and acclimation:

Cirrhinus cirrhosus (Mrigal carp) fingerlings (3.33±0.12 g) were purchased from Fish farm, Thittai, Thanjavur district Tamil Nadu, India and reared in the laboratory in a glass aquarium and acclimated in aerated tap water for two weeks before experiment. During this time, fish were fed a known amount.

Food habits:

Cirrhinus cirrhosis eats from the habitat's intermediate stratum and bottom-feeder (Talwar and Jhingran, 1991). This fish feeds on decaying vegetation, rice bran, wheat bran, mustard oil cake, and other supplementary feed in aquaculture.

Methodology:

In this experiment, fish of uniform length (2.42±0.07 cm) and weight (3.33±0.12 g) were segregated from the stock and acclimated for 3 days to lab conditions, temperature (28±2°C), pH 7.5–7.8, and photoperiod (12:12 h L/D). Ten fish from each of four groups (one control and three experimental) were placed in separate troughs

with dechlorinated tap water. All experimental fish were fed as follow.

Group I: Diet Control,

Group II: 1% Soya Chunk Diet

Group III: 1% Almond Diet II

Group IV: 1 per cent combined diet (1:1 ratio of Diet I and Diet II)

Diet:

Soya Chunk and Almond were purchased from Tamil Nadu, India. The feeds were given to the fingerlings for 30 days. Tanks were cleaned and water changed after every three days. *Growth:*

We measured *Cirrhinus cirrhosis* fingerlings' growth every 15 days for one month. Following formulae were used to measure growth:

$$\text{SGR} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Experimental days}} \times 100$$

$$\text{FCR} = \frac{\text{Feed supplied (dry weight)}}{\text{Body weight growth (wet weight)}}$$

$$\text{Survival (\%)} = \frac{\text{Harvest}}{\text{stock}} \times 100.$$

Estimating composition:

1g tissue was homogenized in 0.1 M Tris HCl buffer (pH 7.4) for biochemical analysis. Protein was estimated by Lowry *et al.* (1951). Anthrone technique was used for carbohydrate analysis. Tissue amino acids was estimated by Rosen's (1957) method.

Statistical Analysis:

The results were analyzed by SPSS Software ver. 20. Significant differences between mean values were evaluated by One Way Analysis of Variance (ANOVA) followed by Duncan's multiple range test (DMRT). Mean values inside columns followed by distinct letters (superscript) are statistically significant ($P < 0.05$) from each other, and same letters are statistically non-significant ($P > 0.05$). One-way ANOVA and post-hoc Tukey HSD were used to examine row mean values. Group I vs. II, III, and IV showed significant variation. * indicates $P < 0.05$ and NS = not significant.

Results and Discussion

Fish need proper nourishment to thrive. Fish devour plants and animals in nature. Artificial feed is needed to sustain a higher fish density than the water's natural fertility can support in semi-intensive fish cultivation (Jhingran, 1991). The nutritional needs of fish depend on the feed used in intensive fish farming. Fish growth rate, feed conversion efficiency, and chemical composition are affected by feed amount and quality (Jena *et al.*, 1998). In this study, Soya Chunk and Almond supplementation was tested on *Cirrhinus cirrhosis* (Mrigal carp) fingerlings.

Soya Chunk, Almond, and Soya Chunk + Almond supplementation on fingerling growth:

Cirrhinus cirrhosis fingerlings with varied feeding regimes were studied. Fingerlings were fed Soya Chunk, Almond, and Soya Chunk + Almond. The fingerlings were fed 1% of their body weight for 15 and 30 days (Table 1).

Tables 2 and 3 show the effect of Soya Chunk, Almond, and Soya Chunk + Almond supplementation in freshwater fish *Cirrhinus cirrhosis* fingerlings (15 and 30 days). After 15 days, fingerlings fed Soya Chunk gained weight (4.79 ± 0.07 g) and length (3.61 ± 0.12 cm) as compared to those fed the control diet (3.34 ± 0.11 g and 2.46 ± 0.11 cm). *Cirrhinus cirrhosis* fingerlings fed Soya Chunk for 30 days gained weight and length (5.15 ± 0.13 g and 3.95 ± 0.11 cm) as compared to those fed the control diet (3.37 ± 0.09 g and 2.49 ± 0.06 cm).

After 15 days, Almond supplementation increased body weight and length in *Cirrhinus cirrhosis* fingerlings (4.23 ± 0.08 g and 3.19 ± 0.10 cm) as compared to the control diet (3.34 ± 0.11 g and 2.46 ± 0.11 cm). Almond supplementation for 30 days increased body weight and length in fingerlings (4.87 ± 0.11 g and 3.65 ± 0.09 cm) as compared to the control diet (3.37 ± 0.09 g and 2.49 ± 0.06 cm).

Cirrhinus cirrhosis fingerlings fed Soya Chunk + Almond for 15 days gained weight and length (5.21 ± 0.13 g and 4.23 ± 0.15 cm) as compared to

those fed the control diet (3.34 ± 0.11 g and 2.46 ± 0.11 cm). *Cirrhinus cirrhosis* fingerlings fed Soya Chunk + Almond for 30 days gained weight and length (5.67 ± 0.12 g and 4.65 ± 0.11 cm) as compared to those fed the control diet (3.37 ± 0.09 g and 2.49 ± 0.06 cm).

Among the various diets and days, combined diet (Soya Chunk + Almond) substantially increased body weight and body length at 30 days as compared to control, other diets and 15 days diet supplementation.

One-way ANOVA and post-hoc Tukey HSD were used to examine row mean values. Group I vs. II, III, and IV showed statistically significant variation. Significance level alpha 0.05. * $P < 0.05$ statistically significant and NS= Non significant ($P > 0.05$) compared with Group I (Control Diet).

Table 4 illustrates the effect of Soya Chunk, Almond and Soya Chunk + Almond supplementation on SGR (%) and FCR of freshwater fish fingerlings with varied feeding regimes (15 and 30 days). After 15 days supplementation of Soya Chunk to *Cirrhinus cirrhosis* fingerlings there was raised SGR (2.44%) and decreased FCR (1.28) as compared with control (SGR 0.01% and FCR 1.51) diet. After 30 days supplementation of Soya Chunk to *Cirrhinus cirrhosis* fingerlings raised SGR (1.46%) and decreased FCR (1.13) were recorded as compared with control (SGR 0.03% and FCR 1.47) diet.

After 15 days supplementation of Almond to *Cirrhinus cirrhosis* fingerlings an increase in SGR (1.55%) and decrease in FCR (1.11) was noticed as compared with control (SGR 0.01% and FCR 1.51). After 30 days supplementation of Almond to *Cirrhinus cirrhosis* fingerlings increased SGR (1.24%) and decreased in FCR (and 0.95) was recorded as compared with control (SGR 0.03% and FCR 1.47) diet.

After 15 days supplementation of Soya Chunk + Almond, *Cirrhinus cirrhosis* fingerlings showed raised SGR (3.04%) and decreased FCR (0.96) as compared with control (SGR 0.01% and FCR 1.51). After 30 days supplementation of Soya Chunk +

Table 1: Ingredients and proximate composition of diets

Ingredients (%)	Control Diet	1% Diet I	1% Diet II	1 % Combined Diet
Fishmeal	35.0	35.0	35.0	35.0
Soybean meal	17.0	17.0	17.0	17.0
Rice bran	11.0	11.0	11.0	11.0
Groundnut oil cake	10.0	10.0	10.0	10.0
Tapioca flour	10.0	10.0	10.0	10.0
Mineral premix	1.5	1.5	1.5	1.5
Vitamin premix	0.5	0.5	0.5	0.5
Wheat flour	15	14	14	14
Soya Chunk	-	1.0	-	0.5
Almond	-	-	1.0	0.5
Proximate composition (%)				
Crude protein	36.13	41.51	39.17	42.69
Crude lipid	7.71	9.73	8.65	9.91
Crude carbohydrate	21.63	25.12	21.18	22.01
Ash	8.15	9.27	9.15	9.31

Table 2: Body weight (g) of *Cirrhinus cirrhosis* fingerling supplemented with formulate diet (Control and Treatment)

Day	Group I (Control Diet)	Group II (1% Diet I)	Group III (1% Diet II)	Group IV (1% Combined Diet)
Initial day	3.33±0.12 ^a	3.32±0.11 ^{a,NS}	3.35±0.09 ^{a,NS}	3.30±0.11 ^{a,NS}
15 th Day	3.34±0.11 ^a	4.79±0.07 ^{b,*}	4.23±0.08 ^{b,*}	5.21±0.13 ^{b,*}
30 th Day	3.37±0.09 ^a	5.15±0.13 ^{c,*}	4.87±0.11 ^{c,*}	5.67±0.12 ^{c,*}

Table 3: Body length (cm) of *Cirrhinus cirrhosis* fingerling supplemented with formulate diet (Control and Treatment)

Day	Group I (Control Diet)	Group II (1% Diet I)	Group III (1% Diet II)	Group IV (1% Combined Diet)
Initial day	2.42±0.07 ^a	2.41±0.11 ^{a,NS}	2.39±0.08 ^{a,NS}	2.43±0.12 ^{a,NS}
15 th Day	2.46±0.11 ^a	3.61±0.12 ^{b,*}	3.19±0.10 ^{b,*}	4.23±0.15 ^{b,*}
30 th Day	2.49±0.06 ^a	3.95±0.11 ^{c,*}	3.65±0.09 ^{c,*}	4.65±0.11 ^{c,*}

Table 4: SGR, FCR and % of Survival of *Cirrhinus cirrhosis* fingerling supplemented with formulate diet (Control and Treatment)

Day	Group I (Control Diet)	Group II (1% Diet I)	Group III (1% Diet II)	Group IV (1% Combined Diet)
Specific growth rate (SGR) % / Day				
15 th Day	0.01	2.44	1.55	3.04
30 th Day	0.03	1.46	1.24	1.80
Food conversion ratio (FCR)				
15 th Day	1.51	1.28	1.11	0.96
30 th Day	1.47	1.13	0.95	0.83
% of Survival				
End of experiment	95	98	98	99

Almond to *Cirrhinus cirrhosus* fingerlings there was increase in SGR (1.80 %), and decrease in FCR (0.83) as compared with control (SGR 0.03% and FCR 1.47) diet.

Supplementation of Soya Chunk, Almond and Soya Chunk + Almond to *Cirrhinus cirrhosus* fingerlings for 30 days caused increase in survival (98%, 98% and 99% for Soya Chunk, Almond and combined diet, respectively) as compared with control (95%) diet.

Among the various diets and days, combined diet (Soya Chunk + Almond) showed substantial increase in SGR, % of survival and lowered FCR at 30 days supplementation as compared to control, other diets and 15 days diet supplementation.

Soya Chunk, Almond, and Soya Chunk + Almond on fingerling composition:

Biochemical parameters of an organism including proteins, lipids, carbohydrates, and amino acids, provide energy for physiological functions. Because different animal tissues and organs are designed to carry out distinct physiological activities, they may have varied organic compositions. Changes in fish carbohydrate, protein, and fat contents may be related to muscle synthesis and deposition rates (Abdel-Tawwab *et al.*, 2006). Biochemical analyses help to assess and control farmed fish health (Ghosh *et al.*, 2003; Cnaani *et al.*, 2004).

The proximate composition of *Cirrhinus cirrhosus* fingerlings with different feeding regimes were examined. Soya Chunk, Almond and Soya Chunk + Almond diet were fed to *Cirrhinus cirrhosus* fingerlings at 1% of their body weight for 15 and 30 days.

Figure 1 shows the effect on carbohydrate (mg/g) of *Cirrhinus cirrhosus* fingerlings after supplementation with Soya Chunk, Almond, and Soya Chunk + Almond. *Cirrhinus cirrhosus* fingerlings showed increased carbohydrate content (1.85 ± 0.05 , 1.45 ± 0.06 and 2.39 ± 0.05 mg/g for Soya Chunk, Almond and combined diet, respectively) after 15 days supplementation with Soya Chunk, Almond and Soya Chunk + Almond

diet as compared with control (1.35 ± 0.05 mg/g) diet. After supplementation with Soya Chunk, Almond and Soya Chunk + Almond for 30 days to *Cirrhinus cirrhosus* fingerlings there was increase in carbohydrate content (2.23 ± 0.06 , 1.78 ± 0.04 and 2.67 ± 0.07 mg/g for Soya Chunk, Almond and combined diet, respectively) as compared to control (1.37 ± 0.07 mg/g) diet. Compared to the control, other diets, and 15-day diet supplementation, the combined diet (Soya Chunk + Almond) had the highest carbohydrate level at 30 days.

Figure 2 depicts the effect of Soya Chunk, Almond and Soya Chunk + Almond supplementation on protein content (mg/g) of freshwater fish *Cirrhinus cirrhosus* (fingerlings). After 15 days feeding with Soya Chunk, Almond, and Soya Chunk + Almond to *Cirrhinus cirrhosus* fingerlings an increased Protein content (5.23 ± 0.05 , 4.51 ± 0.07 , and 5.51 ± 0.09 mg/g for Soya Chunk, Almond, and combined diet, respectively) has been noticed as compared with control (3.57 ± 0.04 mg/g) diet. After 30 days of feeding with combined diet (Soya Chunk + Almond) there was an increased protein content of *Cirrhinus cirrhosus* fingerlings as compared to control, other diets and 15 days diet supplementation.

Figure 3 shows the effect of Soya Chunk, Almond, and Soya Chunk + Almond supplementation on Lipids (mg/g) of freshwater fish *Cirrhinus cirrhosus* (fingerlings). After 15 days, supplementation of Soya Chunk, Almond, and Soya Chunk + Almond to *Cirrhinus cirrhosus* fingerlings increased Lipids content (0.85 ± 0.03 , 0.65 ± 0.01 , and 1.23 ± 0.02 mg/g for Soya Chunk, Almond, and combined diet, respectively) as compared with control (0.42 ± 0.05 mg/g) diet. Among the various diets and days, combined diet (Soya Chunk + Almond) has substantially enhanced lipids content at 30 days as compared to control, other diets and 15 days diet supplementation.

Figure 4 illustrates the effect of Soya Chunk, Almond and Soya Chunk + Almond supplementation on amino acid ($\mu\text{g/g}$) of freshwater fish

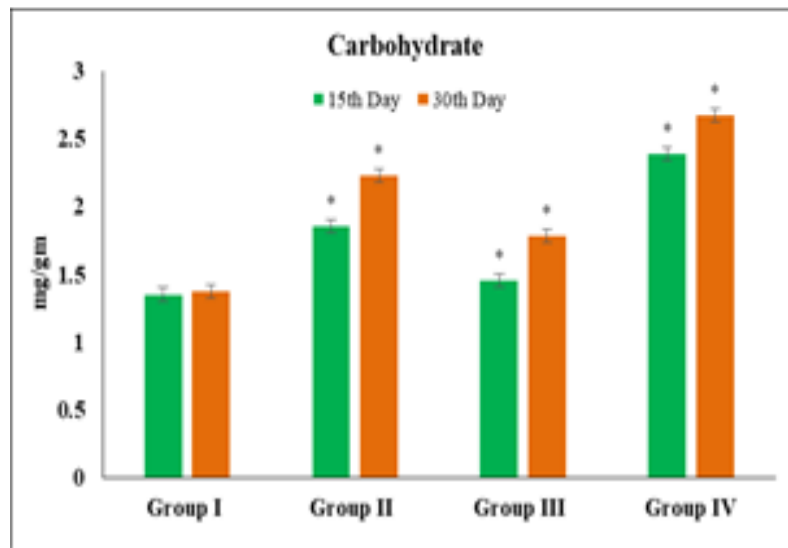


Fig. 1: Carbohydrate (mg/g) content of tissue in *Cirrhinus cirrhosis* fingerling supplemented with formulate diet (Control and Treatment).

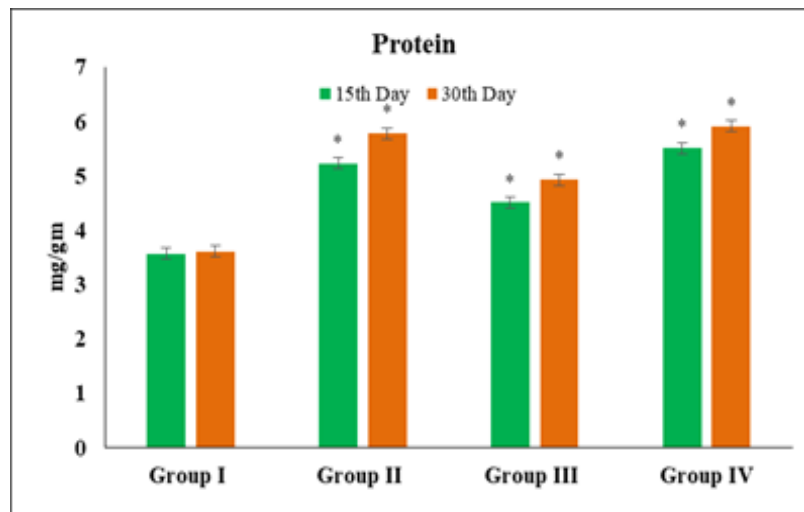


Fig. 2: Protein (mg/g) content of tissue in *Cirrhinus cirrhosis* fingerling supplemented with formulate diet (Control and Treatment).

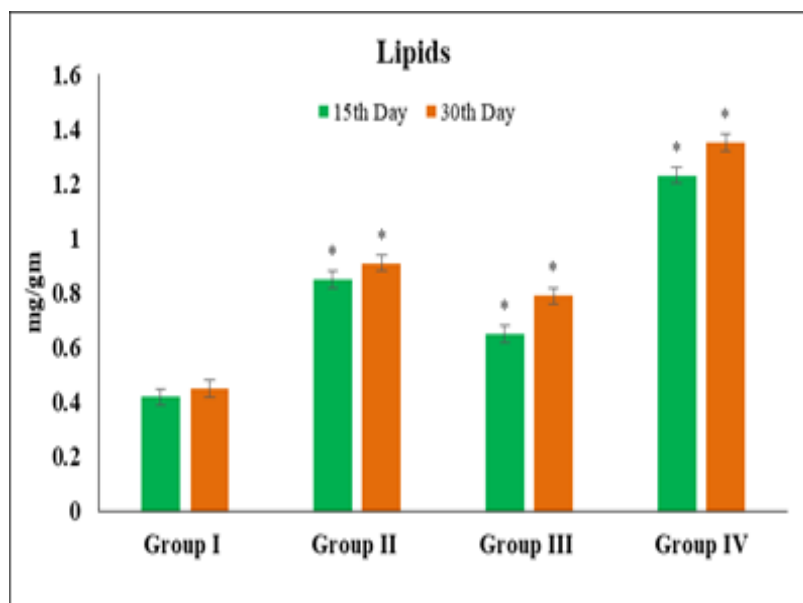


Fig. 3: Lipids (mg/g) content of tissue in *Cirrhinus cirrhosis* fingerling supplemented with formulate diet (Control and Treatment).

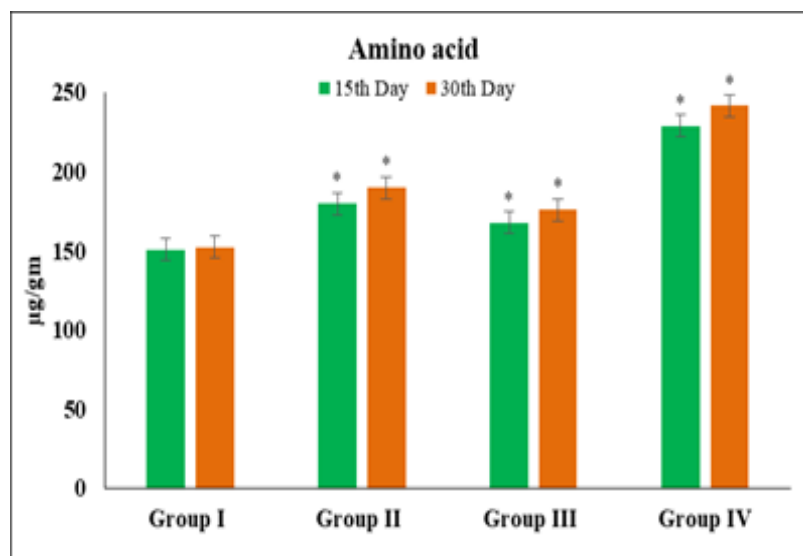


Fig. 4: Amino acid (µg/g) content of tissue in *Cirrhinus cirrhosis* fingerling supplemented with formulate diet (Control and Treatment)

Cirrhinus cirrhosis fingerlings. After 15 days supplementation of Soya Chunk, Almond, and Soya Chunk + Almond to *Cirrhinus cirrhosis* fingerlings there was increased Amino acid content (179.85 ± 2.54 , 167.62 ± 2.73 , and 229.01 ± 2.63 µg/g for Soya Chunk, Almond, and combined diet, respectively) as compared with control (150.57 ± 2.93 µg/g) diet. Compared to the control, other diets, and 15-day diet supplementation, the combined diet (Soya Chunk + Almond) has

significantly higher amino acid content at 30 days.

Carbohydrates are energy precursors for stressed fish (Umminger, 1970). Glucose plays a crucial role in animal bioenergetics, being converted to chemical energy and then mechanical energy. Changes in carbohydrate metabolism, measured as plasma glucose (an energy substrate thought to help animals cope with stress-induced energy demands), are employed as general stress markers in fish (Teles *et al.*, 2007). Glucose (or

glucose 6-phosphate) is released by glycogen phosphorylase (GP) (Roach *et al.*, 1998), and carbohydrate metabolism predominantly supplies energy by oxidising glucose and lactate (Morgan *et al.*, 1997).

Insufficient protein causes stunted growth and weight loss. When too much protein is consumed, only a portion is used for protein synthesis (growth), and the rest is converted to energy (Arredondo *et al.*, 2013). Protein forms the cell membrane and is found inside. Protein forms enzymes, antibodies, hormones, and other bodily regulators. This study found that combination diet-supplemented fish had high protein content. High protein level is due to protein-rich diet. The current investigation confirmed past findings of Storebakken (2000).

Nutritionally, biochemical studies are crucial. Protein is important for life and is the body's most abundant nutrient (Sudhakar *et al.*, 2011). In numerous fish species, proteins are of important as structural components, biocatalysts and hormones for control of growth and differentiations (Amal and Naheb, 2012; Amudha *et al.*, 2017). Protein in fish is a significant component element of tissue and organs. They are precursors to other nitrogen molecules (enzymes, hormones, slurry, neurotransmitters, cofactors, etc.) and provide energy. The influence of dietary protein levels on fish growth performance varies greatly within species, size, age, food and composition, range of proteins level examined and rearing settings (Arredondo, 2013).

Lipids and fatty acids play a major function in membranes and affect membrane-mediated processes like osmoregulation, nutrition absorption, and transport. On the other hand, the nature and quantity of these lipids in fish varies according to species and behaviour. Previous investigations (Kumaran *et al.*, 2012) support our findings.

The glucose concentration was postulated to be mediated by endocrine release such as cortisol (Hontela *et al.*, 1996). Silbergeld (1974) stated

that this parameter can indicate environmental stress. In this study, mixed diet had more carbohydrate than other diets. The higher content of carbohydrate is due to source of carbohydrate content in combination diet. The current investigation derives support from past findings of Gumus and Ikiz (2009).

Conclusion

Supplementing *Cirrhinus cirrhosus* fingerlings with Soya Chunk, Almond, and Soya Chunk + Almond affects growth and biochemical composition. Diets' benefits are related to their length. Among the various diets and days, combined diet (Soya Chunk + Almond) has substantial effect on growth performance and biochemical composition were seen at 30 days supplementation as compared to control, other diets and 15 days diet supplementation. The results of the present study revealed that combination food has superior nutritional supplementation to *Cirrhinus cirrhosus* fingerlings.

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